

Identifying and Revealing Active Compound from Green Tea (*Camellia sinensis*) for Curing Systemic Lupus Erythematosus by Acting as CASPASE 1 Inhibitor

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Abstract

This research aimed to reveal the potency of green tea (*Camellia sinensis*) as immunomodulator, anti-apoptosis agent and anti pyroptosis agent in Systemic Lupus Erythematosus.. The rest samples was downloaded from KNAPSACK Database. Biological activity prediction was conducted by PASS SERVER. Molecular Docking analysis was performed by Autodock Vina in PyRx software v.0.9.3. Complex docking was analyzed and visualized using Discovery Studio Software. The result showed that *Camellia sinensis* has the main potency as immunomodulatory, antioxidant and anti-inflammation. The highest activity of this herbal is antioxidant. There are few bioactive that have role as immunomodulator. The result of molecular docking can explain the mechanism of active compound as anti-apoptosis and anti-pyroptosis. Docking result showed that Theaflavin has the highest affinity to both activities. It can be concluded that bioactive from *Camelia sinensis* is promising for curing Systemic Lupus Erythematosus by acting as antioxidant and inhibit apoptosis and pyroptosis mechanism.

Keywords: *Camellia sinensis*, anti-apoptosis, pyroptosis, Systemic Lupus Erythematosus

Introduction

Systemic Lupus Erythematosus (SLE) is a chronic inflammatory of autoimmune disease where the immune system was mistakenly attacks healthy body cells. The production of autoantibodies damage many organs including the joints, skin, kidneys and nervous system⁽¹⁾. This disease occurs nine times more among women than men. The cause of SLE is unknown, but some occur due to genetic susceptibility⁽²⁾ and environmental factors⁽³⁾ have been showed to be involved in its pathogenesis.

SLE was associated with impaired apoptotic cleansing. While initial triggers were varieties, excessive accumulation of apoptotic debris, especially microparticles containing nuclear material, activate antigen presenting cells including dendritic cells and B cells, which initiate cellular interactions that lead to the generation of antinuclear antibodies through interactions with autoreactive T cells⁽⁴⁾. T cells not only activate B-cell responses but also infiltrate te target tissue and cause type I and type II interferon (IFN I/II) damage, tumor necrosis factors (TNF), B-lymphocyte stimulators (BLys), interleukin 6, interleukin 17, interleukin 18, interleukin 21 and many other cytokines involved in autoimmune priming and inducing inflammation-mediated tissue injury in patients with SLE⁽¹⁾.

Apoptosis in the model of SLE, the nuclear material driving autoreactivity translates to the extracellular space as a consequence of cell death. In this case

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apoptosis as a source of selfantigen in lupus. Another cell death that underlies the pathogenesis of SLE is pyroptosis. Pyroptosis release of nuclear and cellular autoantigens, release of inflammatory cytokines (IL-1b, IL-18) and release of danger molecules (HMGB1, ATP) thus triggering inflammation and inducing autoimmune (5). Caspase activation by the apoptotic pathway results in activation of effector caspase (caspase-3) whereas caspase-1 can cause piroptosis, a form of programmed cell death accompanied by cell lysis (6).

New agents being developed by specifically target the intracellular signaling pathway, inflammatory cytokines, chemokines, surface costimulation molecules, and proteasomes as new drug options in the treatment of SLE. Nowadays, belimumab (anti-BLYS) is the only biologic drug approved by the FDA to treat SLE. Drugs representing various therapeutic strategies are moving into phase III clinical trials (7). New agents with potential as SLE treatment are needed to support the development of SLE therapy.

Another natural therapy as an alternative treatment and prevention of SLE complications is use tea. *Camellia sinensis* tea is classified according to processing used into four different subtypes: green tea, black tea, white tea and oolong tea. Green tea is a product without fermentation process and thus the oxidation of polyphenol components can be prevented. White tea is a product by using minimal fermentation of new shoots and young leaves are harvested only once a year in early spring (8). The making of black tea carried out by fermentation ensures a high level of oxidation of enzymatic catalyzed polyphenols followed by a series of chemical condensation (8-9).

Green tea is beneficial for health such as cancer prevention, obesity, diabetes and neurodegenerative diseases. And some studies showed that the benefits of Green tea for autoimmune diseases such as SLE. All studies proved that the effect of EGCG on autoimmune pathogens such as EGCG preventing and reversing defects in autoimmune encephalomyelitis by inhibiting T cell proliferation by suppressing CDK4 and regulating I κ B- α and directly inhibiting nerve cell death by impairing ROS formation (10). Inhibits autoimmune arthritis by Inhibition of STAT3 and HIF-1a with Th17 / Treg (11). Repairing autoimmune encephalomyelitis by

changing the balance between CD4 T cell subsets by activating MAPKs and NF- κ B signaling pathways (12).

Camellia sinensis tea has many active compounds that have potential as anti-inflammatory, antioxidant and immunomodulatory agents. For this reason, this research explored the potential of tea and green tea (*Camellia sinensis*) as an immunomodulatory, anti-inflammatory, antioxidant and anti-apoptotic and antipyroptosis agent as agents for SLE worship. This research will evaluate using computational process in the early step. Computational analysis or in-silico can reveal the potential of herbal before testing in the laboratory. This technique could explain the mechanism of action by conducting the molecular docking process. *Camellia sinensis* was used for treatment in rheumatoid including Systemic Lupus Erythematosus. *Camellia sinensis* has EGCG bioactive that popular for responsible in many mechanism

Method

Retrieval Data from Database

Bioactive compounds from *Camellia sinensis* were collected from KNAPSACK (<http://kanaya.naist.jp/KNAPSAcK/>). The SMILE format and 3D structure of *Camellia sinensis* of Bioactive was downloaded from PubChem (www.pubchem.ncbi.nlm.nih.gov) (Supplementary Data 1). Moreover, the 3D structure of Caspase 1 and Caspase 3 were downloaded from Protein Data Bank (PDB) (www.rcsb.org/pdb/home/home.do). The 3D structure was provided for molecular docking process

Analysis of Biological Activity

The active compounds of *Camellia sinensis* were analyzed using PASS online server to determine the biological activity. It depended on the compound structure. The analysis will be resulting Pa (Probability of active) and Pi (Probability of inactive) score with range from 0 to 1. If Pa > Pi mean it is potential for specific therapeutic candidate (13).

Molecular Docking

The docking process was carried out using Autodock vina in PyRx 0.9.3 to compute the binding affinity between ligands (active compounds of *Camellia sinensis*) and protein target. Each of active compounds

were docked to specific active site of protein target⁽¹⁴⁾.

Molecular Interaction

The molecular interaction between active compound and protein target was analyzed by Discovery Studio Program. The result determination was based on the amino acid interaction. The active compound would be determining as potential inhibitor if they had interacted to amino acid in the active site of protein target.

Molecular Visualization

All biomolecules were visualized using Discovery Studio to generate the representative figure.

Findings

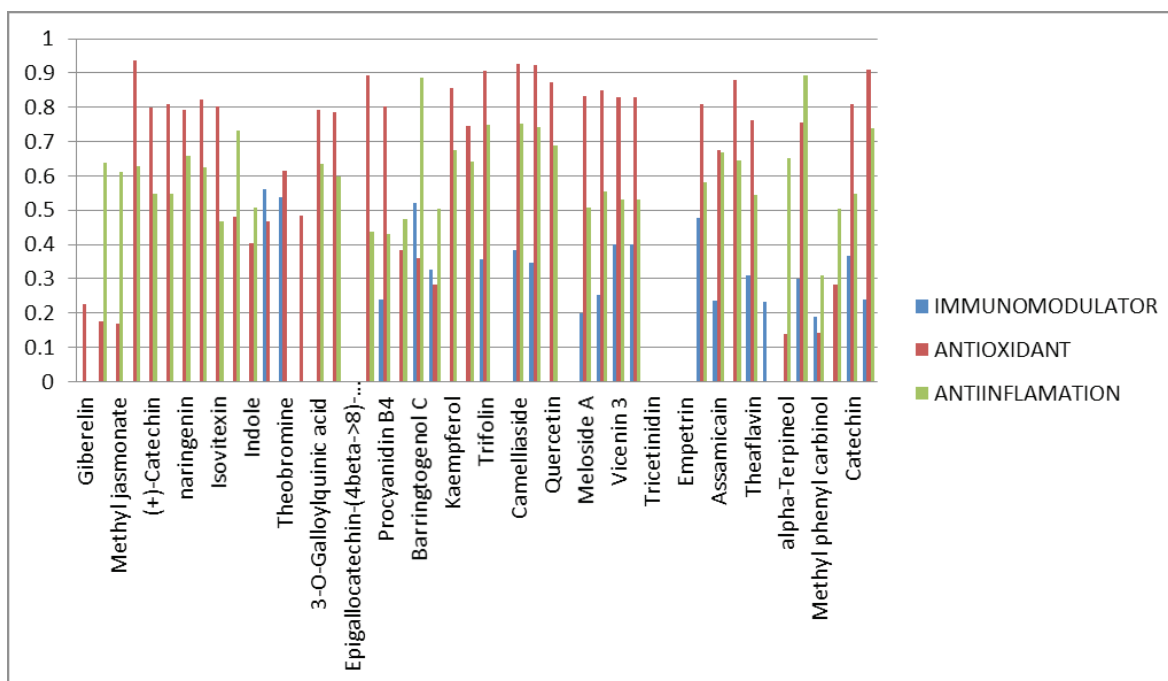


Figure 1. Biological activity prediction of *Camellia sinensis* showed its potentials as immunomodulator, antioxidant, and immunomodulator.

Table 1. Docking Result of *Camellia sinensis* bioactive

No.	Compounds	BINDING AFINITY	
		CASPASE 1	CASPASE 3
1	Cis-Jasmone	-4.6	-5.4
2	Giberelin	-6.2	-6.5
3	Salicylaldehyde	-4.1	-4.7
4	Methyl phenyl carbinol	-4.2	-4.9
5	alpha-Terpineol	-4.3	-5.3
6	Theaflavin	-7.5	-8.7
7	Xanthine	-4.8	-8
8	Idaein	-6.7	-7
9	Empetrin	-6.5	-6.8

Cont... Table 1. Docking Result of *Camellia sinensis* bioactive

10	Tricetinidin	-6	-6.6
11	Isoschaftoside	-6.6	-6.9
12	Vicenin 3	-6.4	-8.2
13	Quercetin	-5.8	-6.8
14	Meloside A	-6.9	-6.6
15	6,8-Di-C-beta-D-arabinopyranosylapigenin	-6.6	-6.6
16	Nicotiflorin	-7	-7.1
17	Camelliaside	-6.1	-6.6
18	Astragalin	-6.7	-6.3
19	Trifolin	-6.8	-6.4
20	Pollenitin	-5.8	-6.6
21	Kaempferol	-5.6	-6.6
22	(E)-Citral	-4.1	-5
23	Barringtogenol C	-6.8	-6.6
24	3-O-Caffeoylquinic acid	-6.1	-7.4
25	Procyanidin B4	-6.6	-6.8
26	Prodelphinidin B4	-6.5	-7.6
27	Theobromine	-4.7	-6.3
28	Theophylline	-4.7	-6.2
29	3-O-Galloylquinic acid	-5.9	-6.7
30	Caffein	-4.7	-5.4
31	Indole	-5.2	-6.4
32	Naringenin	-5.8	-6.9
33	Isovitexin	-6.2	-6.6
34	(-)-Epicatechin	-5.6	-6.7
35	(+)-Catechin	-5.7	-7
36	Dihydroquercetin	-5.7	-7.1

Interaction between theaflavin with caspase 3 and caspase 1 showed that this complexed was stabilized by hydrogen bond and hydrophobic interaction. The active site of Caspase 3 tends to more hydrophobic than caspase 1 (figure 2). **gure 2.** Theaflavin bind to Caspase 1. (figure 2).

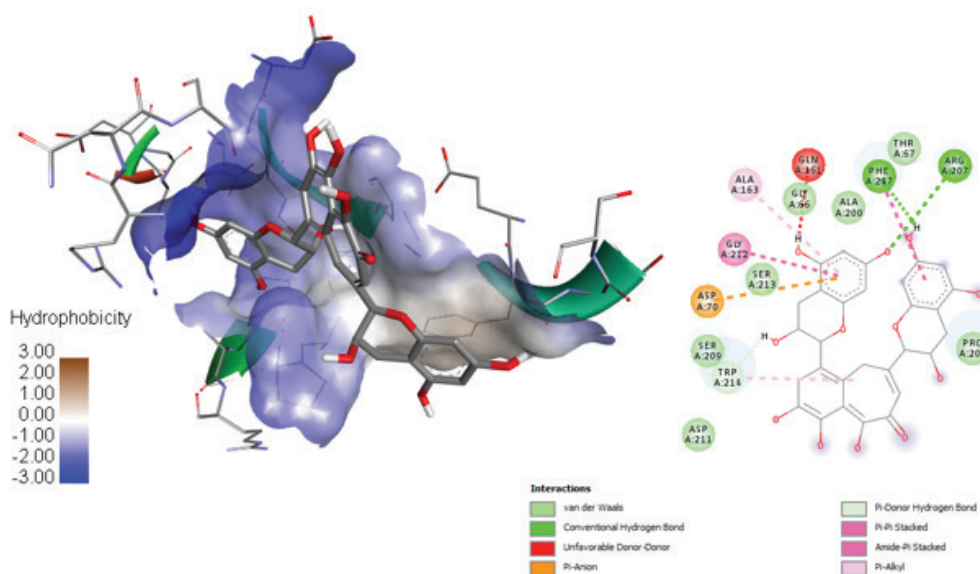


Figure 2. Theaflavin bind to Caspase 1

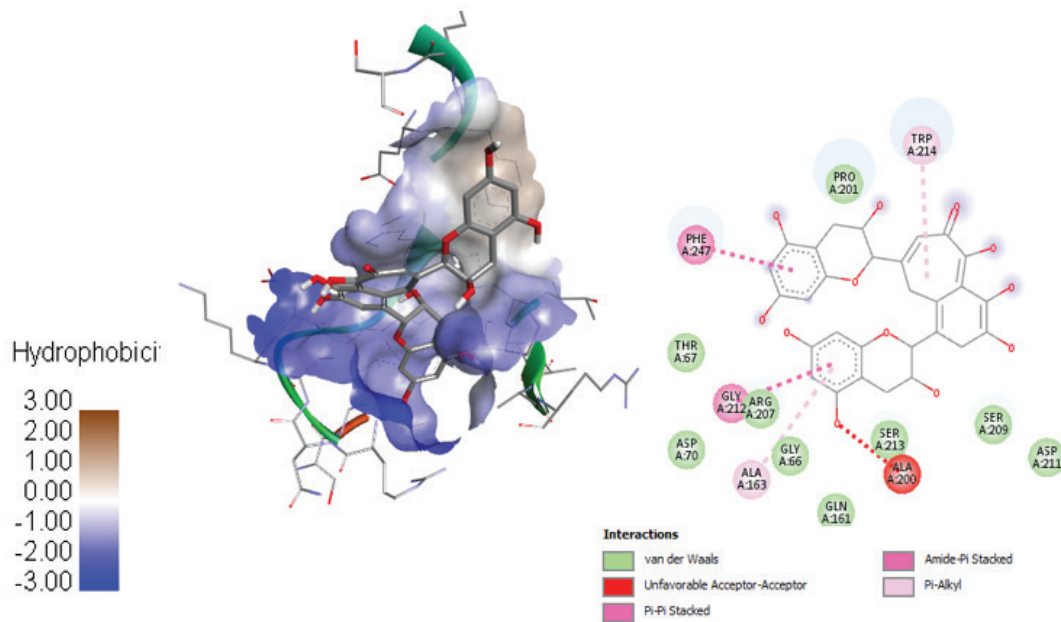


Figure 3. Theaflavin bind to Caspase 3

Discussion

Biological activity prediction of *Camellia sinensis* showed that this herbal has three potentials as immunomodulator, antioxidant, and immunomodulator. Most of compounds have antioxidant activity with the score probability activity (Pa) above 0.7 ($Pa > 0.7$). This result followed by anti-inflammation activity and immunomodulatory activity. Both activities have probability activity above 0.3 ($Pa > 0.3$) (Figure 1). The

value of Pa and Pi can be considered to be measures of the compound to study belonging to active and inactive compounds respectively ⁽¹⁴⁾.

Molecular docking is widely used to identify and predict a new ligand for substrate or protein target in specific site. Generally, the potential of active compound (ligand) is determined by binding affinity value in docking process. Table 1 shows that each of active compound of *Camellia sinensis* has strongly

bind to protein target of Caspase 1 and Caspase 3. The highest affinity is Theaflavin that has binding affinity -7.5 Kcal/mol to Caspase 1 and -8.7 to caspase 3. Apoptosis process is mediated by Caspase 3. To inhibit this mechanism, bioactive compound of *Camellia sinensis* should bind to the active site of Caspase 3. One of bioactive compound from *Camellia sinensis* that promising for inhibitor is theaflavin. Other target is Caspase 1. this protein has the main role in pyroptosis. Surprisingly, the result of molecular docking is theaflavin could bind to this protein as well as caspase 3. The multi-target mechanism of herbal is commonly well-known. This mechanism can reduce the adverse effect and has more benefit than synthetic drug. Systemic Lupus Erythematosus pathogenesis related to innate and adaptive immune response such as inflammasome, apoptosis, clearance apoptosis, pyroptosis (5; 6; 15).

Camellia sinensis can strongly bonded with Caspase 1 as a target for the treatment of systemic Lupus Erythematosus by using the mechanism of Pyroptosis. Pyroptosis is one of the mediator for SLE pathogenesis. Pyroptosis begins an inflammatory process in SLE. The inflammation occurs due to an increasing of immune complexes antinuclear antibodies (ANA) with nuclear antigens. This will triggers the damage of tissue and cell death. Pyroptosis is a form of cell death that occurs SLE. Pyroptosis can be induced by an inflammatory process during the pathogenesis of lupus. Pyroptosis can be characterized by the presence of HMGB1 and ATP where it acts as an inflammatory DAMP. The product of pyroptosis is the release of intact nuclei and the formation of LE cells (5, 16, 17, 18).

Caspase 1 is an enzyme that processes interleukin-1 (IL-1) and IL-18 into its active form during the inflammatory process. In the presence of autoantibodies, the immune complex activates the inflammasome machine in monocytes (19-22). It can be concluded that the precipitating factors for autoimmune if nuclear and cellular autoantigens are found, the release of inflammatory cytokines such as (IL-1 and IL-18) and the release of danger molecules such as HMGB1 and ATP. The changes of immune system during the inflammatory process will trigger to lose of tolerance and induce autoimmune (5, 16, 17, 18).

The process of cell death through pyroptosis or apoptosis will have an impact on tissue damage and immune dysregulation, especially developing into lupus nephritis (23). Considering the process of inflammation and cell death in pathogenesis SLE, a therapy to prevent inflammation and cell death, proven effective in the treatment of SLE.

Conclusion

It can be concluded that *Camellia sinensis* is promising for Systemic Lupus Erythematosus by acting as antioxidant, antiinflammation, antiapoptosis, and anti pyroptosis. Experimental analysis is necessary for proving this result.

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Conflict of Interest-no conflict of interests regarding the publication

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